

CLAIMS

1. An isolated protein, consisting of a protein product of a gene which is structurally related to the *ced-3* and ICE genes, said isolated protein

5 having an alteration in the amino acid sequence of the product of a gene which is structurally related to the *Ced-3* and ICE genes, said alteration corresponding to an alteration in the sequence of SEQ ID NO: 4 selected from the group consisting of:

- i) L to F at amino acid 26;
- ii) G to R at amino acid 65;
- 10 iv) G to S at amino acid 287;
- v) truncation of said protein after amino acid 323;
- vi) truncation of said protein after amino acid 339;
- vii) A to V at amino acid 361;
- viii) E to K at amino acid 390; and
- 15 ix) T to F at amino acid 393.

2. The protease of Claim 1 which cleaves after aspartate residues.

3. The protease of Claim 1 which is a cysteine protease.

4. An isolated ICE polypeptide (SEQ ID NO: 4) having an alteration which reduces the activity of the enzyme, wherein said alteration is selected from the group consisting of:

- 20 a) L to F at amino acid 26;

b) G to R at amino acid 65;

c) G to S at amino acid 287;

d) truncation of said polypeptide after amino acid 323;

e) truncation of said polypeptide after amino acid 339;

5 f) A to V at amino acid 361;

g) E to K at amino acid 390; and

h) T to F at amino acid 393.

5. A product of the gene of Claim 4 selected from RNA and protein.

6. A constitutively activated cell death protein comprising an amino acid sequence, said sequence comprising a portion of the Ced-3 protein shown in SEQ ID NO: 2 of Figure 6A, said portion selected from the group consisting of:

a) the amino acids from approximately 150 to 503 (SEQ ID NO: 20);

b) the amino acids from approximately 373 to 503 (SEQ ID NO: 21);
and

15 c) the amino acids from approximately 150 to 372 (SEQ ID NO: 22).

7. The constitutively activated cell death protein of claim 6, further comprising a subportion of the region of Ced-3 from amino acids 1 to 149, as shown in SEQ ID NO: 2 of Figure 6A, said subportion enhancing the activity of
20 the protein.

8. A constitutively activated cell death protein having an amino acid sequence ICE from the sequence shown in Figure 6A (SEQ ID NO: 4), said sequence selected from the group consisting of:

5 a) the amino acids from approximately 111 to 404 (SEQ ID NO: 23);
 b) the amino acids from approximately 298 to 404 (SEQ ID NO: 24);
 c) the amino acids from approximately 111 to 297 (SEQ ID NO: 25).

9. An isolated protein which is the NEDD-2 protein (SEQ ID NO: 26) having an alteration which inactivates the protein, wherein said alteration is A to V at amino acid 117.

10 10. The isolated protein of claim 9, wherein said alteration is C to A at amino acid 303.

11. The isolated protein of claim 9, wherein said alteration is C to S at amino acid 303.

15 12. Isolated protein which is selected from the group consisting of Ced-3 (SEQ ID NO: 2), ICE (SEQ ID NO: 4), and NEDD-2 (SEQ ID NO: 13), said protein having an alteration at a conserved amino acid corresponding to an amino acid of the Ced-3 protein (SEQ ID NO. 2) selected from the group consisting of:

20 a) Ced-3 Ser 183 or ICE Ser 126;
 b) Ced-3 Met 234;
 c) Ced-3 Arg 242;
 d) Ced-3 Leu 246 or ICE Leu 166;

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- e) Ced-3 Ile 247 or ICE Ile 167;
- f) Ced-3 Ile 248 or ICE Ile 168;
- 5 g) Ced-3 Asn 250 or ICE Asn 170;
- h) Ced-3 Phe 253 or ICE Phe 173;
- i) Ced-3 Arg 259 or ICE Arg 179;
- j) Ced-3 Gly 261 or ICE Gly 181;
- k) Ced-3 Asp 265 or ICE Asp 185;
- 10 l) Ced-3 Gly 277 or ICE Gly 197;
- m) Ced-3 Tyr 278 or ICE Tyr 198;
- n) Ced-3 Val 280 or ICE Val 200;
- o) Ced-3 Lys 283 or ICE Lys 203;
- p) Ced-3 Asn 285 or ICE Asn 205;
- 15 q) Ced-3 Leu 286 or ICE Leu 206;
- r) Ced-3 Thr 287 or ICE Thr 207;
- s) Ced-3 Met 291 or ICE Met 211;
- t) Ced-3 Phe 298 or ICE Phe 218;
- 20 u) Ced-3 His 304 or ICE His 224;
- v) Ced-3 Asp 306 or ICE Asp 228;
- w) Ced-3 Ser 307, ICE Ser 229, or NEDD-2 Ser 16;
- x) Ced-3 Leu 310, ICE Leu 232, or NEDD-2 Val 19;
- y) Ced-3 Val 311, or ICE Val 233, or NEDD-2 Val 20;
- 25 z) Ced-3 Ser 314 or ICE Ser 236;
- aa) Ced-3 His 315 or ICE His 237;
- bb) Ced-3 Gly 316 or ICE Gly 238;
- cc) Ced-3 Ile 321, ICE Ile 243, or NEDD-2 Leu 23;
- dd) Ced-3 Gly 323, ICE Gly 245, or NEDD-2 Asp 25;

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- ee) Ced-3 Ile 334, ICE Ile 261, or NEDD-2 Phe 31;
- ff) Ced-3 Asn 339 or ICE Asn 266;
- gg) Ced-3 Pro 344 or ICE Pro 271;
- hh) Ced-3 Leu 346 or ICE Leu 273;
- ii) Ced-3 Lys 349 or ICE Lys 276;
- jj) Ced-3 Pro 350, ICE Pro 277, or NEDD-2 Pro 37;
- kk) Ced-3 Lys 351 or ICE Lys 278;
- ll) Ced-3 Gln 356, ICE Gln 283, or NEDD-2 Glu 43;
- mm) Ced-3 Ala 357, ICE Ala 284, or NEDD-2 Thr 44;
- nn) Ced-3 Cys 358 or ICE Cys 285;
- oo) Ced-3 Arg 359, ICE Arg 286 or NEDD-2 Arg 46;
- pp) Ced-3 Gly 360, ICE Gly 287, or NEDD-2 Gly 47;
- qq) Ced-3 Asp 371 or ICE Asp 297;
- rr) Ced-3 Asp 414, ICE Asp 326, or NEDD-2 Asp 82;
- ss) Ced-3 Arg 429, ICE Arg 341, or NEDD-2 Arg 97;
- tt) Ced-3 Gly 434, ICE Gly 346, or NEDD-2 Gly 102;
- uu) Ced-3 Ser 435, ICE Ser 347, or NEDD-2 Ser 103;
- vv) Ced-3 Ile 438, ICE Ile 350; NEDD-2 Ile 106;
- ww) Ced-3 Ala 449, ICE Ala 361, or NEDD-2 Ala 108;
- xx) Ced-3 Val 454, ICE Val 366, or NEDD-2 Val 123;
- yy) Ced-3 Leu 488, ICE Leu 394, or NEDD-2 Leu 158;
- zz) Ced-3 Tyr 493, ICE Tyr 399, or NEDD-2 Tyr 163; and
- aaa) Ced-3 Pro 496, ICE Pro 402, or NEDD-2 Pro 166.

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13. The isolated protein selected from the group consisting of Ced-3 (SEQ ID NO: 2) and ICE (SEQ ID NO: 4), said protein having an amino acid alteration in an amino acid corresponding to Cys 358 of Ced-3, and Cys 285 of ICE.

14. The isolated protein of claim 13, wherein said alteration is a Cys to Ala
5 alteration.

15. The isolated protein of claim 13, wherein said protein is ICE and said alteration is at conserved amino acid 285 of said ICE.

16. The isolated protein of claim 13, wherein said protein is NEDD-2 and said alteration is at conserved amino acid 303 of said NEDD-2.

10 17. Isolated nucleic acid encoding the protein of Claim 12.

18. A method of preventing cell death, said method comprising administering a polypeptide of claim 12.

19. The method of claim 14, wherein said administering is to a patient and said polypeptide is provided at a therapeutically effective dose.

15 20. A method of preventing cell death, said method comprising administering a therapeutically effective amount of the isolated nucleic acid of claim 17.